Mini-review: diabetic renal complications, a potential stinky remedy

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Sen U, Pushpakumar S. Mini-review: diabetic renal complications, a potential stinky remedy. Am J Physiol Renal Physiol 310:F119–F122, 2016. First published November 4, 2015; doi:10.1152/ajprenal.00299.2015.—Chronic kidney disease is associated with vasculitis and is also an independent risk factor for peripheral vascular and coronary artery disease in diabetic patients. Despite optimal management, a significant number of patients progress toward end-stage renal disease (ESRD), a suggestion that the disease mechanism is far from clear. A reduction in hydrogen sulfide (H2S) has been suggested to play a vital role in diabetic vascular complications including diabetic nephropathy (DN). This mini-review highlights the recent findings on the role of H2S in mitigating abnormal extracellular matrix metabolism in DN. A discussion on the development of the newer slow-releasing H2S compounds and its therapeutic potential is also included.

diabetic nephropathy; hydrogen sulfide; NMDA receptor; remodeling; signaling

DIABETIC NEPHROPATHY (DN) is one of the leading causes of end-stage renal disease (ESRD) throughout the world. Despite advances in understanding the disease process, the precise mechanism of initiation and progression remains unclear. In addition to high glucose, diabetes is associated with other metabolic derangements including protein (9, 30), lipid (35) and gaseous molecules such as, nitric oxide (NO) (41) and hydrogen sulfide (H2S) (45). Current evidence suggests that the pathogenesis of DN is multifactorial, and hyperglycemia mediates injury by several mechanisms such as fructokinase activation and ATP depletion, oxidative stress, production of inflammatory cytokines, activation of fibroblasts, and microaneurysm formation (12, 19, 36). Furthermore, at the cellular and molecular level an imbalance of matrix metalloproteinases and their inhibitors leads to abnormal extracellular matrix (ECM) metabolism (8, 27), and disrupted gap junction proteins cause poor cell-cell communication (43). A deficiency of nitric oxide has been implicated in advanced diabetic nephropathy (34). In recent studies, reduction of H2S-producing enzymes and plasma H2S has been associated with chronic kidney disease and diabetic nephropathy (2, 49). In light of current literature, this mini-review will focus on H2S biology and its role as a modulator and potential therapeutic agent in DN.

H2S Production in the Diabetic Kidney

In the last two decades, H2S has overcome its past reputation as a toxic gas and gained attention as a molecule involved in several biological functions. H2S was initially described as a neuromodulator by Abe and Kimura in 1996 (1). In the last decade, other studies have described its role in vasorelaxation (48), angiogenesis (4), nociception (28, 42), cytoprotection (15, 40), myocardial ischemia-reperfusion injury (7), atherosclerosis (5), including diabetic complications (25, 29). Endogenously, H2S is generated by cystathionine β-synthase (CBS), cystathionine γ-lyase (CSE), 3-mercaptoppyruvate sulfurtransferase (3MST) together with cysteine amino transferase (CAT), and D-amino acid oxidase (DAO) in concert with cystathionine β-lyase (CSE), 3-mercaptopyruvate sulfurtransferase (3MST) and cysteine amino transferase (CAT) and D-amino acid oxidase (DAO). A detailed pathway of H2S synthesis has recently been reviewed by Beltowski (3).

Although the distribution of H2S-producing enzymes is tissue specific, the kidney is known to express CBS and CSE (14). In diabetes, the level and activity of both these enzymes are reported to be impaired, and thus H2S production leads to endothelial and other cell type injury (18, 21, 44). A lower plasma H2S concentration in DN patients on hemodialysis was shown to correlate with the progression of atherosclerosis (23). Other researchers including our own study showed that H2S production in the diabetic-induced (44, 49) or type-1 diabetic kidney (17, 18) is decreased.

Role of H2S in Diabetic Kidney Remodeling and Function

Excess matrix protein synthesis and deposition occur in the diabetic kidney. Recent reports suggest that the H2S-producing enzymes and its levels are decreased in diabetes. Upon endogenous induction or exogenous supplementation of H2S, matrix remodeling was mitigated, suggesting a correlation between H2S deficiency and matrix accumulation. In streptozotocin-induced diabetic rats, H2S therapy improved renal function, decreased glomerular basement thickening, mesangial expansion, and interstitial fibrosis (49). In the same study, the authors documented that the reduction of glucose-induced oxidative stress by H2S was mediated by activation of the Nrf2 antioxidant pathway that exerts an anti-inflammatory effect by inhibiting NF-κB signaling in in vitro podocytes (49). This finding is in line with our own study on the genetic type-1 diabetic model, where plasma H2S levels were low and associated with increased extracellular matrix deposition and reduced vascular compliance which was mitigated by H2S treatment (17).

Previously, our laboratory demonstrated that matrix metalloproteinase-9 (MMP-9) diminishes H2S production in the...
type-1 diabetic (Akita) model and this, in part, by reducing CBS and CSE enzyme expression (18). Similar results were also observed in high-glucose treatment in in vitro experiments using glomerular endothelial cells (18). The regulation of CBS/CSE enzymes by MMP-9 was confirmed using Akita and MMP-9 knockout mice along with partial mitigation of renal remodeling in double knockout (DKO) mice (17). In addition, we demonstrated that H2S therapy improves renal function (17).

**Signaling Cross Talk Between Gaseous Modulators in the Diabetic Kidney**

Recently, Lee et al. (20) reported that H2S inhibited high glucose-induced protein synthesis in renal epithelial cells (21). In a separate study, the same group also reported tadalafil, a phosphodiesterase 5 inhibitor, abrogated high glucose-induced global protein synthesis and matrix protein laminin-γ by increasing the expression and activity of H2S-producing enzyme CSE (20). They also observed that in podocytes, tadalafil-induced AMP-activated protein kinase (AMPK) phosphorylation was mitigated by CSE inhibitor DL-propargylglycine and small interfering RNA against CSE in podocytes (20). The authors concluded that high-glucose-induced matrix protein synthesis in podocytes involved a complex interaction of the nitric oxide (NO)-H2S-AMPK axis (20). To further define molecular mechanisms of H2S signaling in diabetic conditions, we performed in vitro experiments using mouse glomerular endothelial cells. The cells were stimulated with high-glucose and treated without or with a H2S donor. Our results suggested that high glucose induced markers of matrix accumulation and autophagy through a LKB1/STRAD/MO25 dependent pathway (Fig. 1) (16).

**The N-Methyl-D-Aspartate Receptor, H2S, and Diabetic Nephropathy**

Although H2S does not have a specific receptor, it has been reported to modulate the N-methyl-D-aspartate (NMDA) receptor in the brain (13). The NMDA receptor is a glutamate receptor and ion channel protein mainly found in the nerve cells involved in controlling synaptic plasticity and memory function (22). In 2002, Deng et al. (6) demonstrated that NMDA receptors were also expressed in the kidney cortex and exerted a tonic vasodilatory response. Inhibition of the receptors caused marked renal vasoconstriction and reduction in renal blood flow in response to glycine infusion (6). However, it was not known whether H2S affects the NMDA receptor in the kidney to modulate function. For the first time, our laboratory demonstrated the role of H2S in renal NMDA receptor-mediated remodeling and dysfunction (18). Our findings suggested that diabetic kidney remodeling was, in part, due to reduced H2S production due to MMP-9-mediated inhibition of CBS and CSE enzyme activity. (18). This was associated with disruption of gap junction proteins, connexin-40 and -43. The above changes were reversed by exogenous H2S supplementation (18). More recently, we demonstrated that an increased renal-resistive index (RI), excess ECM deposition, elevated plasma creatinine, and diminished renal vascular density and cortical blood flow were normalized with H2S treatment in Akita kidneys (17). These findings suggest that H2S has the potential to prevent diabetic nephropathy by preserving renal microvascular architecture and function. A possible pathway of H2S-dependent renal remodeling and signaling mechanism is shown in Fig. 1.

**Perspective and Concluding Remarks**

In summary, studies on H2S have revealed a significant role in a variety of physiological and pathophysiological processes. A decrease in H2S level has been implicated in several pathologies, including diabetic nephropathy, in animals and humans (11, 47, 49). In contrast, increased H2S levels have been reported to contribute to β-cell apoptosis, leading to reduced mass and thus insulin, neutrophil infiltration, and inflammation in lipopolysaccharide-induced endotoxic shock in the lung and liver and synovial inflammation (24, 39, 46). However, the measurement of H2S in some of the earlier studies is inaccurate and overestimated due to the lack of sensitive techniques and partly due to its volatile nature. In addition, studies which showed improvement in pathology used crude H2S donors such
as sodium hydrogen sulfide (NaHS), which is not suitable for human therapy due to its short-time bioavailability and possible toxicity. Current research is now focused on developing novel H_{2}S-releasing compounds, such as GYY4137, AP39, and SG-1002, which are known to simulate the endogenous production of H_{2}S in the body. These compounds are safer than bolus doses of NaHS or sodium sulfide (Na2S), which instantly releases H_{2}S. Furthermore, the slow-releasing compounds can deliver H_{2}S over a longer period of time. In animal models, GYY4137 has been shown to improve vasculopathy (26, 31). Since supplementation of H_{2}S has the potential to prevent and/or improve DN by preserving renal microvasculature and function, additional experiments are needed to test the safety and efficacy using the newer H_{2}S-releasing compounds. In early phase I and phase II clinical trials, the novel H_{2}S donor SG-1002 significantly increased blood levels of H_{2}S and nitrite, suggesting increased nitric oxide availability in healthy and heart failure subjects (32, 33). To fully exploit the therapeutic potential of H_{2}S, larger multicenter cohort studies are required to investigate its beneficial effects not only in the diabetic kidney but also in the other organs affected by diabetes.

Recently, a clinical trial using N-acetyl cysteine (NAC), a derivative of cysteine and substrate for H_{2}S, has completed in patients with chronic kidney disease and patients on dialysis (ClinicalTrials.gov Identifier: NCT01232257). The objective of this study was to examine whether NAC treatment would increase plasma H_{2}S levels and decrease oxidative stress and inflammation. While the results are awaited, it can be speculated that the outcomes will likely provide us insight into the H_{2}S backup mechanism during nitric oxide deficiency in chronic kidney disease patients.

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REFERENCES
Review

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